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Research article

Paleoparasitological analysis of human remains from a European cemetery of the 17th–19th century in Rio de Janeiro, Brazil

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ABSTRACT

Most paleoparasitological studies based on archeological sites in the New World are from pre-Columbian times. However, understanding of the introduction and spread of parasites with the arrival of European settlers and African slaves in America remains a topic for investigation. This study evaluated the presence of intestinal parasites in human remains from an archeological site of the colonial period, and compared the sensitivity of three parasitological techniques for paleoparasitological study. Samples were collected from the archeological site *Nossa Senhora do Carmo* Church, Rio de Janeiro, Brazil. Paleoparasitological examination revealed intestinal helminths in 2/17 (11.8%) individuals. *Trichuris trichiura* and *Ascaris* sp. eggs were found. The spontaneous sedimentation technique showed a greater numerical recovery of parasites, while the flotation techniques were superior in retrieving more parasite types. The study demonstrated that combining the three techniques improves the recovery of parasites in terms of number and diversity. Similar diversity of parasites to that of a previous historical archeological site suggests that the distribution of intestinal parasites was widespread in Rio de Janeiro, regardless of social status.

1. Introduction

Paleoparasitological study is becoming more widespread among researchers in paleopathology. The identification of parasites in human remains has generated information about infectious diseases and lifestyle of ancient populations, as well as helping to chart human migration routes (Araújo et al., 2008; Ferreira, 2011).

Most paleoparasitological studies in the New World have been focused on pre-Columbian times (Ferreira et al., 1980; Araújo et al., 1981; Iñiguez et al., 2003). However, the introduction and spread of parasites after the arrival of European settlers and African slaves to America remains understudied. Evidence of *Trichuris trichiura* and trichostrongylid nematode infections have been reported in archeological sites from the colonial period of Brazil, in the Minas Gerais and Piauí states (Confalonieri et al., 1981; Araújo et al., 1984). Recently, Jaeger et al. (2013) found infections with the nematodes *Trichuris* sp., *Ascaris* sp., and the cestode *Taenia* sp. in individuals buried in a cemetery in Rio de Janeiro dating to the 18th century.

The city of Rio de Janeiro, on the Atlantic coast, has a semi-humid tropical climate with an annual rainfall in the range 1000–1500 mm. The *Nossa Senhora do Carmo* Church (INSC) is located in the neighborhood of the *Largo do Paço*, in the center of Rio de Janeiro. Historically, the church was designated the Chapel Royal, and later it became the cathedral of the city. In 2007, during an architectural and artistic restoration, an archeological site was identified by the Institute of Brazilian Archeology [Instituto de Arqueologia Brasileira (IAB)]. Forty-three human burials and ossuaries were identified, dating from the 17th to 19th centuries (Dias, 2008). Bio-anthropological analysis revealed that most of the subjects were adults less than 30 years old (78%), with an equal number of men and women (Jaeger et al., 2012). A tuberculosis paleogenetic study conducted in the INSC population showed positivity for bacteria of the *Mycobacterium tuberculosis* complex in 53.1% of the individuals (Jaeger et al., 2012). Using the mitochondrial DNA, European ancestry was identified in 100% of primary burials. Considering that intestinal parasite infection is an indicator of health and hygiene conditions of a population, the aim of this study was to evaluate the presence of intestinal parasites in samples from the archeological site of the *Nossa Senhora do Carmo* Church. In addition, the sensitivity of three parasitological techniques in paleoparasitological analyses was compared.

2. Materials and methods

Sediment samples were collected directly from the pelvic region of the seventeen human bodies. All samples were derived

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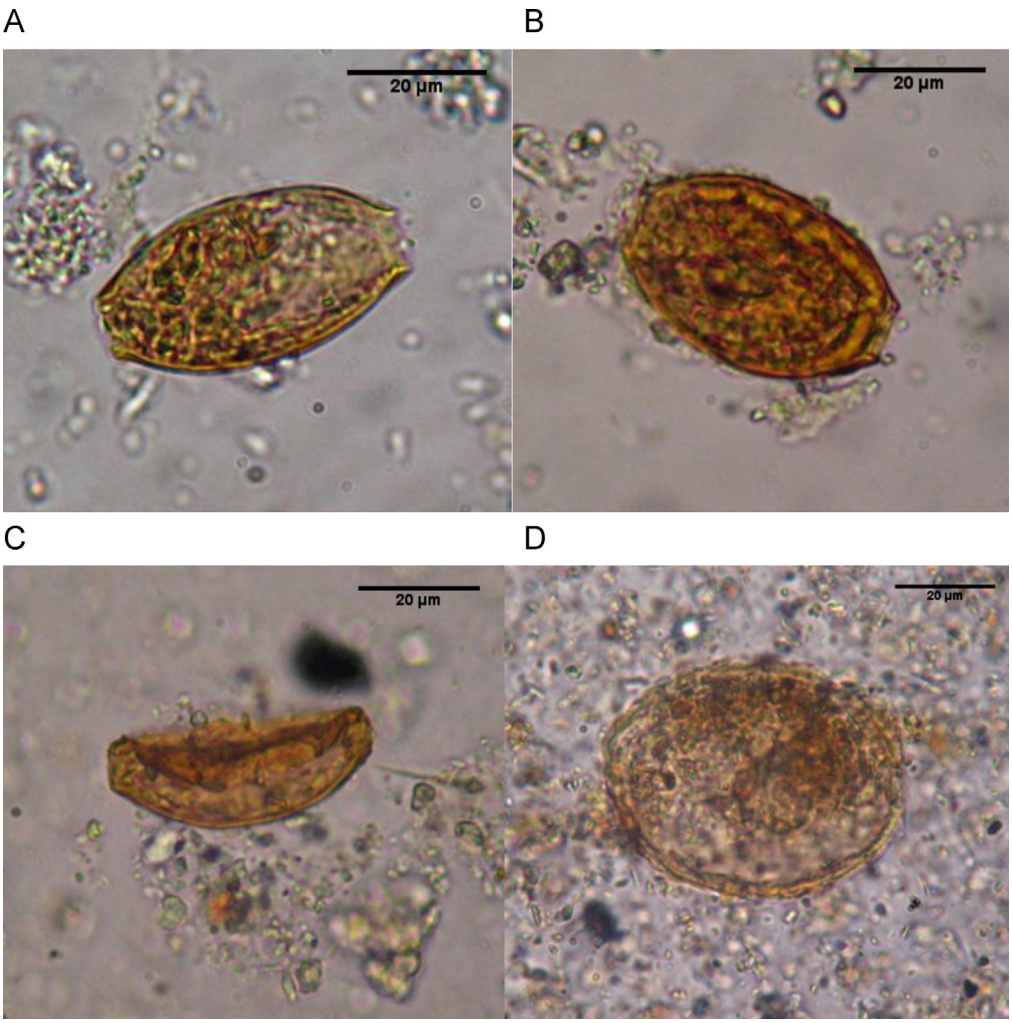

Fig. 1. Paleoparasitological results of archeological site INSC, Rio de Janeiro, Brazil. (A, B) *Trichuris* sp. eggs; (C) warped *Trichuris* sp. egg; (D) *Ascaris* sp. egg. Scale bar in the figures.

from primary burials, with the exception of burial 38, which was from secondary burial (Jaeger et al., 2012). The samples (1–2 g) were rehydrated (1:2, w/v) with 0.5% aqueous trisodium phosphate at 4 °C for 72 h. Paleoparasitological investigation was conducted using three techniques, which followed the protocol of Taglioretti et al. (2012): spontaneous sedimentation, sucrose flotation, and zinc chloride flotation. We conducted the three parasitological techniques on all samples in an effort to obtain the maximal results and for comparative purposes.

For spontaneous sedimentation (Lutz, 1919), ten slides of each sample were examined. Sucrose flotation was conducted at a sucrose density of 1.2 (Sheather, 1923, modified). At least 3 slides were examined after 15, 45, and 105 min flotation or until no further structures were recovered. The sediment samples were cleaned twice with aqueous trisodium phosphate 0.5%, and zinc chloride flotation was conducted at a density of 1.5 (Reinhard et al., 1988). At least 5 slides were examined after 10, 25, 40, 55, and 70 min flotation or until no structures were observed. A minimum of 18 slides was examined for each sample. Slides were examined under 100× and 400× magnification, and measured with an ocular micrometer. The images were made by Canon AS 650, and edited by ImageJ 1.44p (National Institutes of Health, USA). The program R (version 2.13.2) was used for descriptive statistical analyses and construction histograms.

3. Results

When the results of all three methods are combines, we find that two of seventeen (11.8%) individuals had intestinal helminthes (Table 1). A single positive sample was found when using spontaneous sedimentation (5.9%) with 10 of *Trichuris* sp. eggs recovered. Sucrose flotation and zinc chloride flotation each showed a single (5.9%) positive sample for *Ascaris* sp. and *Trichuris* sp., respectively (Table 1).

Two methods (spontaneous sedimentation and zinc chloride flotation) recovered at total of 13 *Trichuris* sp. eggs from individual 14A (Table 1 and Fig. 1A–C). Most *Trichuris* sp. eggs possessed no content or polar plugs. *Trichuris* sp. egg deformation ($n = 3$) was observed with zinc chloride flotation (Fig. 1C), and data from those samples were excluded from the descriptive analysis. *Trichuris* sp. eggs ($n = 10$) measured 40.0–47.5 µm in length and 22.5–27.5 µm in width (mean ± SD = 43.5 ± 2.48 µm L and 24.0 ± 1.84 µm W) (Fig. 2). The *Trichuris* sp. mean egg size corresponds to that established for *T. trichiura* (Confalonieri et al., 1988). A single *Ascaris* sp. egg measuring 57.5 × 45.0 µm was found in individual 38H (Fig. 1D), using the sucrose flotation technique.

4. Discussion

In the Americas, parasite infections became a major health problem after the arrival of European settlers and the slave trade.